Attorney's Docket No.: 13028-002001 / P12999

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant : Anke Rattenholl et al. Art Unit : 1647
Serial No. : 09/807,096 Examiner : Robert C. Hayes
Filed : November 19, 2001
Title : METHOD FOR OBTAINING ACTIVE BETA-NGF

Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

DECLARATION BY DR. SUSAN LOREY UNDER 37 C.F.R. 1.132

I, **Susan Lorey**, hereby declare that:

1. I conducted or supervised the following two experiments, in which β-NGF and proNGF were compared for their ability to induce migration of (1) murine fibroblasts, and (2) bovine corneal endothelial cells.

In organisms cellular migration is of importance in all processes where cells have to be recruited to places they are required, e.g. migration of inflammatory cells, fibroblasts, endothelial or epithelial cells to support wound healing. *In vitro* cellular migration may be studied in the Boyden Chamber as model for examining cell movement through a porous membrane towards a chemoattractant.

The proNGF and β-NGF were generated as described in the Specification of US 09/807,096, e.g., at page 13, line 14 through page 25, line 26.

2. In a first experiment, embryonic murine fibroblasts (3T3) were used to study the effect of proNGF and β-NGF on the migration of these cells. Cellular migration of the cells stimulated

CERTIFICATE OF MAILING BY FIRST CLASS MAIL

I hereby certify under 37 CFR §1.8(a) that this correspondence is being deposited with the United States Postal Service as first class mail with sufficient postage on the date indicated below and is addressed to the Commissioner for Patents, P.O. Box 1450, Alexandria, VA 22313-1450.

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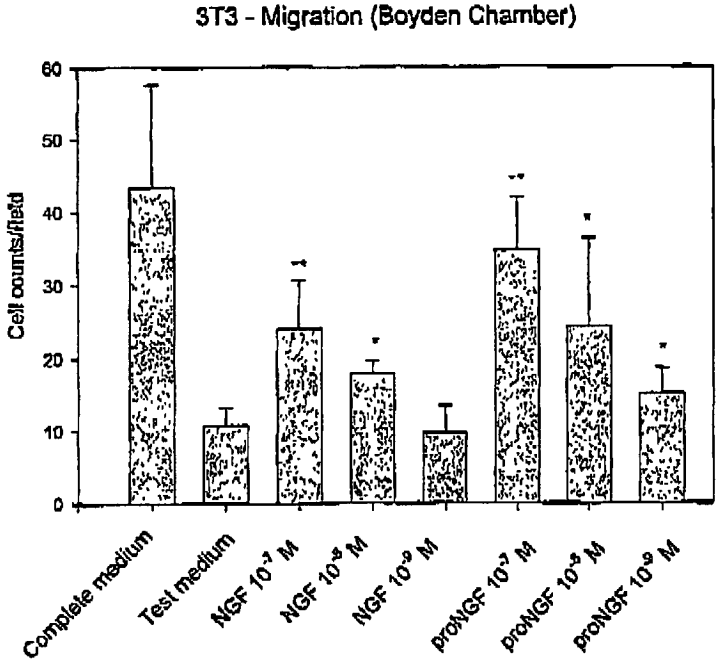
by proNGF, β-NGF or a test medium negative control was determined in a Boyden Chamber after a period of 4 hours.

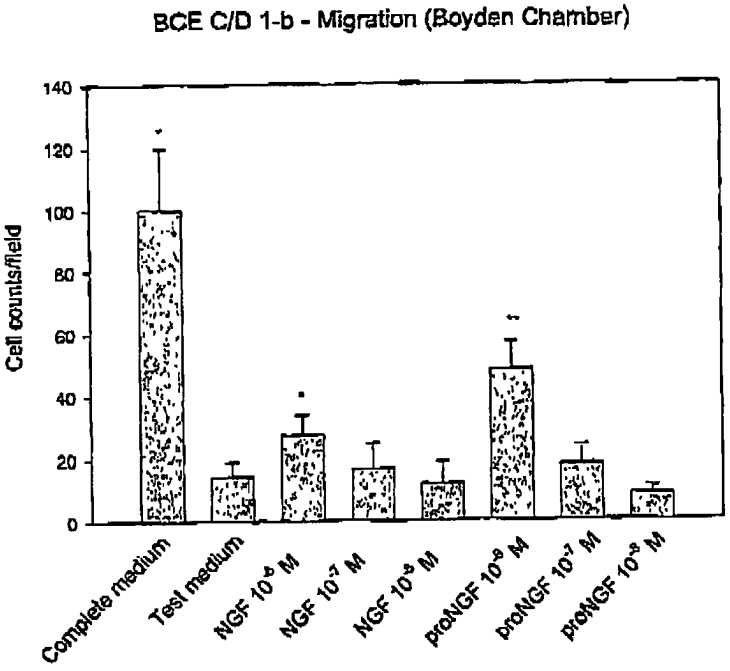
At all doses, proNGF had significantly greater efficacy than β-NGF in inducing murine fibroblast migration, as compared to the negative control.



3. In a second experiment, bovine corneal endothelial cells were used to study the stimulatory effects of proNGF and β-NGF on the migration of these cells. In a Boyden Chamber, corneal endothelial cells were stimulated with proNGF, β-NGF, or a test medium negative control. Cellular migration was determined after a period of 4 hours.

ProNGF (10^{-6} M) had significantly greater efficacy than β-NGF (10^{-6} M) in inducing bovine epithelial cell migration, as compared to the negative control.

Based on these results, it was concluded that proNGF itself acts as an active ingredient. In support of this conclusion, it is noted that if proNGF exhibited its activity only after cellular cleavage of the proNGF into β-NGF, proNGF could not have greater activity than β-NGF as observed in these experiments.



4. All statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

Respectfully Submitted,

Date: 22. 12. 2005

i.A. Susan Lorey

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